Effects of Honey Bee (Hymenoptera: Apidae) Foraging on Seed Set in Self-fertile Sunflowers (*Helianthus annuus* L.)

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ABSTRACT Ten self-fertile commercial sunflowers cultivars were evaluated for seed set with and without exposure to bees. In the first planting, the number of foraging honey bees was smaller than in the second, and seed set for most cultivars did not differ between those bagged to exclude bees and ones that were open pollinated. In the second planting, however, a majority of cultivars had significantly greater seed set when capitula were exposed to bees compared with when they were not. The weight of seeds from open-pollinated capitula was greater than from those where bees were excluded. Environmental conditions also played a role in seed set as evidenced by differences between plantings in set on bagged capitula. In the first planting, average maximum and minimum temperatures were significantly higher than in the second, and overall seed set was significantly lower in capitula where bees were excluded compared with the second planting. Under the high temperature conditions, however, some cultivars set four times more seed on open-pollinated capitula compared with those that were bagged. These results suggest that foraging activity and cross-pollination by bees might mitigate reductions in seed set caused by high temperatures.

KEY WORDS *Apis mellifera*, *Helianthus annuus*, pollen viability, stigma receptivity, temperature, humidity

The importance of honey bees in the production of sunflower [*Helianthus annuus* L. variety *macrocarpus* (D.C.) (Ckll)] seed has been the subject of numerous studies (Free 1964, Parker 1981, Fell 1986, Skinner 1987, DeGrandi-Hoffman and Martin 1993). Bees certainly are essential in seed production for male-sterile (i.e., nonpollen producing) sunflowers because pollen must be transferred from male-fertile to male-sterile plants. However, the importance of bees for seed set in self-fertile sunflowers where male and female florets exist on the same flower head (capitulum) is less clear. Some studies provide evidence that more seed is set when bees forage on sunflowers (Parker 1981, Krause and Wilson 1981, dePaiva et al. 2003), but there also are reports that because hybrid sunflower cultivars are self-fertile, bees are not required for seed set or provide only a slight increase in seed yields (Myers 2002).

Sunflowers are highly attractive to many species of bees, but the primary pollinator of commercial plantings is the honey bee (McGregor 1976, Parker 1981). A capitula is composed of hundreds or thousands of individual florets. When florets first open, they are protandrous, but by the second day, the stigma pushes up from the base of the floret. Any pollen left on the anthers could contact the stigma resulting in self-pollination. In self-fertile cultivars, self-pollination can produce seed set. Bees also pollinate florets while they move among them collecting nectar and pollen.

Although hybrid sunflowers are self-fertile, the degree to which they are self-pollinating is uncertain. For self-pollination to lead to seed set, viable pollen must contact the stigma at a time when it is receptive. The optimum time for self-pollination would be when the stigma is pushing up through the anthers. However, if the timing of either pollen availability or stigma receptivity is not synchronous, seed set will not occur.

Whether pollination leads to seed set also is dependent on environmental factors, particularly temperature and relative humidity. Limitations on seed set caused by the effects of temperature (>30°C) on pollen germination and pollen tube growth have been documented in plants such as cotton (*Gossypium hirsutum*) (Reddy et al. 1992, Kakani et al. 2005), corn (*Zea mays*) (Mitchell and Petolino 1988), and tomato (*Lycopersicon esculentum*) (Peet et al. 1998, Sato et al. 2002). High temperatures at preanthesis can cause pollen sterility, and at anthesis, retarded pollen tube growth (Prasad et al. 2001). High temperatures coupled with the drying effects of low relative humidity also can affect female floral structures causing reductions in the duration of stigma receptivity, pollen germination on the stigmatic surface, and initial pollen tube growth (Hedhly et al. 2004, Ortega and Dicenta 2004). High temperatures also can accelerate ovule

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degeneration (Postweiler et al. 1985, Cerovic et al. 2000).

Whether seed production in self-fertile sunflowers is affected by weather conditions has not been studied, nor has whether the effects can be mitigated by bee pollination. The purpose of this study was to compare seed yields between capitula exposed to bees and those where bees were excluded in 10 commercial self-fertile sunflower cultivars. The effects of environmental factors on set were explored by repeating the studies under conditions where bloom period temperatures differed.

Materials and Methods

Description of the Study Site. All data were collected at the University of Arizona Agricultural Research Facility in Tucson, AZ, in 2004. Uncultivated desert surrounding the Facility and a dry river bed provided nesting sites for non-*Apis* bees. The Carl Hayden Bee Research Center Apiary (with >70 colonies) was <1.0 km from the study site, thus insuring adequate populations of honey bees.

Ten commercial self-fertile sunflower cultivars were used in the study. The cultivars were 'Interstate HYB-6767', 'Interstate HYSUN 450 AK10', 'Interstate HYSUN 525 HYBSF', 'Mycogen SF-260', 'Mycogen 8D 310', 'Mycogen 8N429CL', 'Mycogen 8N421', 'SunRea Hybrid', 'Legend LSF-124N', and 'Legend LSF-146N'. The cultivars were chosen because they are commonly planted self-pollinating commercial cultivars. Each cultivar had a single capitulum per plant and protandrous florets. The cultivars were planted in groups of three rows. The 30 rows used for data collection had border rows on either side. Data were not collected from the border rows. The arrangement of cultivars was randomized separately for each planting. Rows were 96.9 m long. The first planting began blooming on 22 June and continued until 2 July. Bloom on the second planting began on 4 October and continued until 18 October. Weather data were collected hourly at the AZMET monitoring station established at the site (http://ag.arizona.edu/azmet).

Counts of *Apis* and non-*Apis* Pollinators. Honey bees and non-*Apis* bees were counted on each cultivar by walking up and down rows in the morning and again in the afternoon throughout bloom and counting the number of bees actively collecting nectar or pollen (voucher specimens of honey bees and non-*Apis* bees counted on sunflowers are maintained at the Carl Hayden Bee Research Center, Tucson, AZ). One row was counted per cultivar for each observation period. Two observers counted bees simultaneously in different rows. It took ≈2–3 min to count the bees in each row (depending on foraging activity); therefore, each bee count on all cultivars took ≈10–15 min. After the bee counts were completed, the number of capitula with open florets in the rows was counted so that data were expressed as bees per capitulum. Bee counts began for each cultivar when at least 10% of the flowers in a row were open and attracting bees. We continued the counts daily until the cultivar was no longer attractive to bees except when weather was not conducive for flight or the field was being irrigated.

Observations of Self-Pollination and Estimates of Seed Set. The average percentage of florets that set seed in the field was compared between capitula that were bagged in a nylon mesh so that bees could not forage on them (hereafter referred to as bagged) and those that were not bagged and were foraged by bees throughout bloom (open). The bag completely enclosed the capitulum so that neither pollen nor nectar was accessible to bees. The bags remained on the capitula until all florets had senesced after which the plant was tagged for identification as a bagged capitula. Ten bagged and 10 open capitula were sampled for each cultivar to estimate seed set in each planting. The capitula were sampled when the seeds were black and ready to harvest. In the second planting, seed weight was also compared between bagged and open capitula. Twenty-five seeds from eight capitula were weighed for each cultivar. As in the seed set estimates, capitula were sampled when the seeds were black and ready to harvest.

To be certain that the cultivars could self-pollinate, we cut sunflowers from the field plots and placed them in the laboratory. The capitula (five from each cultivar) were selected at random from each cultivar when <1% of the florets were opened. The stems were put in flasks filled with water. The capitula were not bagged or exposed to bees and were left undisturbed until ≈70% of the florets opened. The stigma on the capitula were examined for pollen by examining them under a dissecting microscope (Leica MZ12; Leica Microsystems, Bannockburn, IL)

Statistical Analysis. Average daily maximum and minimum temperatures, high and low relative humidity, and heat units per day were compared between plantings using *t*-tests. Heat units were estimated using the single sine method (Arnold 1960, Allen 1976). Temperature ranges for heat unit accumulation were 10–30°C, which are the lower and upper thresholds for honey bee foraging activity, nectar secretion, and anther dehiscence (Ribbands 1953, Free 1970).

Two-way analysis of variance (ANOVA) was used to determine the effects of cultivar and planting (factors) on honey bee foraging activity (response variable) (Sokal and Rohlf 1995). ANOVAs were used to determine if honey bee foraging activity differed among the cultivars in each planting. Comparisons were made between the average numbers of honey bees foraging on each cultivar during the two plantings using *t*-tests.

Average seed set from bagged and open capitula of each cultivar were compared using a three-way ANOVA with planting, cultivar, and bagged versus open capitula as factors and seed set (percentage of florets setting seed) as the response variable. Two-way ANOVA was used to determine the effects of cultivar and open versus bagged capitula on seed weight. One-way ANOVAs were used to compare average seed set and weight from bagged and open capitula among the cultivars within a planting. Seed set from

Table 1. Results of two-way ANOVA comparing honey bee foraging activity on 10 commercial self-fertile sunflower cultivars in two plantings in Arizona

Factor	df	*F*	*P*
Cultivar	9	4.57	<0.0001
Planting-1 versus planting-2	1	9.67	0.002
Cultivar × planting	9	3.09	0.002

bagged and open capitula of each cultivar were compared using *t*-tests.

Results

Weather Conditions. In planting-1, average daily maximum and minimum temperatures ranged between 19.1 and 37.3°C for the 10-d bloom period, whereas in planting-2, where bloom lasted for 14 d, the averages were 12.0–30.8°C. The temperature ranges were significantly different between the two plantings (maximum: $t = 7.0$, $P < 0.00001$, df = 19; minimum: $t = 6.6$, $P < 0.00001$, df = 16). The hours of sunlight were greater in the first planting (14.05 ± 0.003 h) than in the second (11.2 ± 0.02 h), causing daytime temperatures to remain high longer. The differences in daylength and temperatures are reflected in the average number of accumulated heat units per day, which were significantly different between the two plantings (planting-1 = 29.1, planting-2 = 20.1; $t = 8.62$, $P < 0.0000$, df = 23). Relative humidity also differed between the plantings. In planting-1, the average daily ranges of relative humidity were 8.2–49.7%, whereas in planting-2 they were 13.6–66.0% (low: t = −2.16, $P = 0.04$, df = 22; high: t = −3.8, $P = 0.001$, df = 19).

Counts of Honey Bees and non-*Apis* Bees. The bee population was composed primarily of honey bees and some non-*Apis* bees. The non-*Apis* we counted most often were *Bombus*, *Halictus*, and *Mellisodes* species. The average number of honey bees per capitulum on all cultivars was significantly greater than non-*Apis* in both plantings (planting-1: 0.28 ± 0.01 honey bees and 0.005 ± 0.0005 non-*Apis*; $t = 20.6$, df = 119, $P < 0.00001$; planting-2: 0.38 ± 0.03 honey bees and 0.003 ± 0.001 non-*Apis*, $t = 8.91$, df = 58, $P < 0.00001$). Because of the low number of non-*Apis* bees in both plantings, there was no further analysis of non-*Apis* foraging data.

The average number of honey bees per capitulum was significantly affected by the cultivar and planting and the interaction between them (Table 1). Foraging activity did not differ among the cultivars in the first planting, but in planting-2, there were significant differences (Fig. 1). 'Interstate HYSUN-450AK10' had the highest average number of honey bees per capitulum, and 'Interstate HYB-6767' and 'Legend LSF-124-N' had the lowest. There were significantly more foraging honey bees counted on all cultivars combined in planting-2 compared with planting-1 ($t_{96} = -2.80$, $P = 0.006$).

Observations of Self-pollination and Estimates of Seed Set. We examined stigma on capitula of each cultivar that were placed in flasks with water in the laboratory. Pollen grains were present on all stigmas. The capitula did not remain viable long enough for seed to set. However, pollen contacted the stigma in all cultivars in the absence of pollinators or any physical disturbance indicating that all cultivars could self-pollinate.

The average percentage of florets setting seed was significantly affected by planting, cultivar, open or bagged capitula, and the interaction between planting and whether capitula were open or bagged (Table 2). Average set on all cultivars combined was significantly higher in open versus bagged capitula in both plantings (planting-1: open pollinated = 50.7 ± 2.3%, bagged = 35.9 ± 2.1%; $t_{155} = -4.82$, $P < 0.0001$; planting-2: open-pollinated = 71.5 ± 1.6%, bagged = 45.8 ± 1.9%; $t_{149} = 9.88$, $P < 0.0001$).

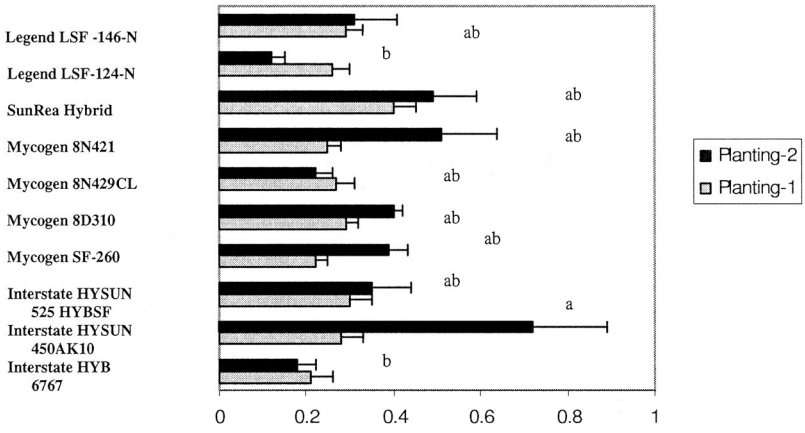

Fig. 1. Average number of honey bees per capitula on self-fertile sunflower cultivars in Arizona. Means were estimated from daily counts of honey bees taken in the morning and afternoon on all cultivars. Means from planting-2 followed by the same letter are not significantly different at the 0.05 level as determined by a Tukey's W procedure (critical value = 0.135) (Sokal and Rohlf 1995). There was no significant difference in honey bee foraging activity among the cultivars in planting-1 ($F_{9,65} = 3.53$, $P = 0.10$).

Table 2. Results of a three-way ANOVA comparing seed set on 10 commercial self-fertile sunflower cultivars in two plantings in Arizona

Factor	df	*F*	*P*
Cultivar	9	4.51	<0.0001
Planting	1	52.42	<0.0001
Open versus bagged	1	52.63	<0.0001
Cultivar × planting	9	1.52	0.142
Cultivar × open versus bagged	9	1.48	0.155
Planting × open versus bagged	1	73.94	<0.0001

Capitula were either open so that bees could pollinate them or bagged to exclude bees.

The percentage of florets that set seed on open capitula was greater than in those that were bagged for three cultivars in the first planting ('Interstate HYB-6767', 'Mycogen 8N421', and 'Legend LSF-146N') and six cultivars in the second ('Interstate HYSUN 450AK10', 'Mycogen SF 260', '8D310', '8N429CL', 'SunRea Hybrid', and 'Legend LSF-124N') (Table 3). In planting-1, 'Interstate HYB-6767' set fewer seeds on bagged capitula compared with all other cultivars except 'Interstate HYSUN 450AK10'. In planting-2, set was significantly lower on 'SunRea Hybrid' and 'Legend LSF-124N' compared with 'Interstate HYB 6767', 'Interstate HYSUN 525 HYBSF', and 'Legend LSF-146N' when capitula were bagged. There was no significant difference in set from open capitula for either planting.

Overall, seed set was higher in planting-2 compared with planting-1 in both open ($t = -7.73$, $P < 0.00001$, df = 145) and bagged capitula ($t_{152} = -3.79$, $P = 0.0002$). Seed set from open capitula differed between plantings for 'Interstate HYSUN 450AK10' ($t_{12} = -3.6$, $P = 0.004$), 'Mycogen 8N429CL' ($t_8 = -3.25$, $P = 0.01$), 'Mycogen 8D310' ($t_{13} = -5.07$, $P = 0.0002$), 'SunRea Hybrid' ($t_8 = -2.46$, $P = 0.04$), 'Legend LSF-124N' ($t_{13} = -3.83$, $P = 0.002$), and 'Legend LSF-146N' ($t_{14} = -3.46$, $P = 0.004$). Seed set from most cultivars did not differ between the two plantings for bagged capitula with the exception of 'Interstate HYB 6767' ($t = -5.75$, $P = 0.003$, df = 9), 'Interstate HYSUN 450AK10' ($t_{13} = -2.45$, $P = 0.029$), and 'Legend LSF-146N' ($t_{14} = -3.46$, $P = 0.004$). There was no difference in seed set between plantings or between open and bagged capitula for 'Interstate HYSUN 525 HYBSF' (bagged: $t_{12} = -1.72$, $P = 0.12$; open: $t_9 = -1.00$, $P = 0.34$; planting-1: $t_{10} = -1.38$, $P = 0.2$; planting 2: $t_9 = 0.68$, $P = 0.519$).

Seeds from open capitula were larger than those that were bagged. Average seed weight per cultivar was significantly affected by cultivar, open or bagged capitula, and the interactions between the two factors (Table 4). Across all cultivars, the average weight of 25 seeds from open pollinated capitula was significantly greater than from those that were bagged (open pollinated = 0.96 g, bagged = 0.62 g; $t_{144} = 10.4$, $P < 0.00001$). With the exception of 'Interstate HYB-6767' and 'Mycogen 8N429CL', the seeds from the individual cultivars weighed more when the capitula were exposed to bees (Table 3). Seed from open-pollinated 'Interstate HYSUN 525 HYBSF' and 'Legend LSF

Table 3. Average percentage of florets setting seed on 10 self-fertile sunflower cultivars in Arizona

Cultivar	Percent florets setting seed				Weight of 25 seeds (g)	
	Planting-1		Planting-2			
	Bagged (*n*)	Open (*n*)	Bagged (*n*)	Open (*n*)	Bagged (*n*)	Open (*n*)
Interstate HYB-6767	11.3b (8)	58.5[a] (8)	54.6a (8)	69.5 (8)	0.8a (8)	0.9b (8)
Interstate HYSUN 450 AK10	32.4ab (8)	40.6 (8)	44.9ab (8)	68.3[a] (8)	0.6ab (8)	1.1ab[a] (8)
Interstate HYSUN 525 HYBSF	41.5a (8)	55.1 (8)	58.2a (8)	64.7 (8)	0.6ab (8)	0.8bc[a] (8)
Mycogen SF-260	51.0a (7)	50.9 (8)	43.8ab (8)	72.2[a] (8)	0.4b (8)	1.0b[a] (8)
Mycogen 8D310	37.7a (7)	46.1 (8)	42.5ab (8)	76.4[a] (8)	0.5b (8)	1.0b[a] (8)
Mycogen 8N429CL	43.5a (8)	44.8 (8)	46.5ab (8)	80.7[a] (8)	0.6ab (8)	0.7c (8)
Mycogen 8N421	44.2a (8)	61.1[a] (8)	48.9ab (10)	64.6 (8)	0.8a (8)	1.1ab[a] (8)
SunRea Hybrid	33.8a (10)	51.5 (8)	35.6b (8)	78.3[a] (8)	0.7a	0.8bc[a]
Legend LSF-124N	32.4ab (7)	36.2 (8)	28.4b (8)	62.1[a] (8)	0.7a (8)	1.3a[a] (8)
Legend LSF–146N	35.5a (7)	56.4[a] (8)	65.1a (9)	77.9 (8)	0.5b (8)	0.7c[a] (8)
Overall	35.9	50.7[a]	45.8	71.5[a]	0.6	0.9[a]

Capitula were either open and could be foraged by bees or bagged and inaccessible to bees.

Means followed by the same letter within a column are not significantly different at the 0.05 level as determined by a Tukey's pairwise comparisons test (Sokal and Rohlf 1995). Percent seed set, bagged: planting-1; critical value = 25.7, planting-2; critical value = 25.0. Weight of seeds: open; critical value = 0.23, bagged; critical value = 0.22.

[a] Means are significantly different between bagged and open capitula at the 0.05 level for the cultivar as determined by a *t*-test.

Table 4. Results of two-way ANOVA comparing seed weight of 10 commercial self-fertile sunflower cultivars in planting-2 in Arizona

Factor	df	*F*	*P*
Cultivar	9	10.38	<0.0001
Bagged versus open	1	220.13	<0.0001
Cultivar × bagged versus open	9	9.74	<0.0001

Capitula were either open so bees could pollinate them or bagged to exclude bees.

124N' weighed the most, and 'Interstate HYB 6767', 'Mycogen SF-260', '8D310', '8N429CL', 'Sun Rea Hybrid', and 'Legend LSF-146N' weighed the least. 'Interstate HYB-6767', 'Mycogen 8N421', 'SunRea Hybrid', and 'Legend LSF-124N' seeds weighed the most, and 'Mycogen SF-260' weighed the least when capitula were bagged (Table 3).

Discussion

The cultivars used in this study were self-fertile and self-pollinating, but there was an overall increase in seed set and in seed weight when capitula were exposed to bees. There was higher seed set overall in the second planting compared with the first possibly because of lower temperatures, higher humidity, and greater numbers of honey bees per capitulum. The low number of non-*Apis* bees in both plantings probably limited their effect on seed set. Evidence for this is that there were larger numbers of non-*Apis* bees counted in planting-1 compared with planting-2, but overall seed set was greater in the second planting compared with the first. There also were differences between the two plantings in seed set of bagged capitula, indicating that high temperatures and low humidity played a role in determining seed set.

Increases in seed set when capitula were available to honey bees could have been caused by both increased self-pollination and possibly cross-pollination. Bees transfer pollen among florets on the same capitulum while they forage and also frequently transfer pollen between rows. In studies with male-sterile sunflowers, honey bees foraging on male-sterile rows often had pollen on their bodies from male-fertile capitula located up to six rows away (DeGrandi-Hoffman and Martin 1995). In our study, all cultivars could self-pollinate. If sufficient amounts of self-pollen were deposited on stigma in the absence of pollinators, seed set should have been similar between bagged and open capitula. Indeed, in the first planting, seed set between open and bagged capitula was the same in most cultivars, but this may have been caused by a combination of low honey bee populations, high temperatures, and low relative humidity. In the second planting, however, there were larger honey bee populations and lower temperatures with higher humidity and most cultivars set more seed when open pollinated. Those four cultivars ('Interstate HYB-6767', 'HYSUN 525 HYBSF', 'Mycogen 8N421', and 'Legend LSF-146N') that did not differ in seed set between open and bagged capitula probably were the most self-compatible and the best at self-pollinating of the 10 we tested, because unlike the other cultivars, seed set was optimized without pollination by foraging bees. However, yields from all but 'Interstate HYB 6767' still could be increased by bee foraging because seed weight was greater on open-pollinated capitula compared with bagged.

The greater number of honey bees counted in planting-2 compared with planting-1 could have been caused by temperature differences between the plantings. Plants will decrease the amount of nectar they secrete when temperatures are high, and thus the florets are less attractive to bees (Ribbands 1953). Although honey bee foraging activity was greater on some cultivars in planting-2, seed set did not differ among open-pollinated capitula. These results indicate that the differences in foraging activity among cultivars probably were not great enough to affect seed set.

Average high temperatures during the bloom period of planting-1 were within the ranges where pollen germination and stigma receptivity can be reduced (Rosell et al. 1999, Ortega et al. 2004). The differences in seed set from bagged capitula among the cultivars could be indicative of their heat tolerance. Those cultivars that set the most seed in the first planting and whose seed set did not differ between plantings could be the most heat tolerant (e.g., 'Interstate HYSUN 525 HYBSF') of those we tested. Conversely, cultivars that set more seed on open capitula compared with bagged in the first planting could be ones whose yields would benefit most by introducing honey bee colonies to increase foraging populations.

Differences in seed set on bagged and open capitula between plantings show how interactions between environment and bee foraging activity influence seed set. If only self-pollination occurred, as in the bagged capitula, either the loss of pollen viability or stigma receptivity would prevent seed set. In the first planting, 'Interstate HYB-6767' and 'Legend LSF 146N' had lower percentages of seed set on bagged capitula compared with the second planting, indicating that temperature might have affected pollen viability or germination. However, open pollinated capitula in those cultivars set more seed than ones that were bagged in the first planting, indicating that pollination by bees might have mitigated the limitations on seed set because of environmental conditions.

Yields from some self-fertile cultivars can be increased in both seed number and weight if bee populations are present during bloom. For cultivars that are less heat tolerant or not completely self-fertile, bees might improve set by transferring viable compatible pollen on to stigmas while they are still receptive. Our field had 10 cultivars, but commercial plantings might have only 1. However, the movement of pollen by foraging bees might also improve set in large fields planted with a single self-fertile cultivar if pollen viability was reduced in particular sections of the field because of microenvironmental factors.

References Cited

Allen, J. C. 1976. A modified sine wave method for calculating degree days. Environ. Entomol. 5: 388–396.

Arnold, C. Y. 1960. Maximum and minimum temperatures as a basis for computing heat units. Proc. Am. Soc. Hortic. Sci. 76: 682–692.

Cerovic, R., D. Ruzic, and N. Micic. 2000. Viability of plum ovules at different temperatures. Ann. Appl. Biol. 137: 53–59.

DeGrandi-Hoffman, G., and J. H. Martin. 1993. The size and distribution of the honeybee (*Apis mellifera* L.) cross-pollinating population on male-sterile sunflowers (*Helianthus annuus* L.). J. Apic. Res. 32: 135–142.

DeGrandi-Hoffman, G. and J. H. Martin. 1995. Does a honey bee (*Apis mellifera*) colony's foraging population on male-fertile sunflowers (*Helianthus annuus* L.) affect the amount of pollen on nestmates foraging on male-steriles? J. Apic. Res. 34: 109–114.

dePaiva, G. J., Y. Terada, and V. de Alencar Arnaut de Toledo. 2003. Seed production and germination of sunflower (*Helianthus annuus* L.) in three pollination systems. Acta Sci. Anim. Sci. 25: 223–227.

Fell, R. D. 1986. Foraging behaviors of *Apis mellifera* L. and *Bombus spp.* on oilseed sunflower (*Helianthus annuus* L.). J. Kansas Entomol. Soc. 59: 72–81.

Free, J. B. 1964. The behaviour of honeybees on sunflowers (*Helianthus annuus* L.). J. Appl. Ecol 1: 19–27.

Free, J. B. 1970. Insect pollination of crops. Academic, New York.

Hedhly, A., J. I. Hormaza, and M. Herrero. 2004. Effect of temperature on pollen tube kinetics and dynamics in sweet cherry, *Prunus avium* (Rosaceae). Am. J. Bot. 91: 558–564.

Kakani, V. G., K. R. Reddy, S. Koti, T. P. Wallace, P.V.V. Prasad, V. R. Ready, and D. Zhao. 2005. Differences in *in vitro* pollen germination and pollen tube growth of cotton cultivars in response to high temperature. Ann. Botany 96: 59–67.

Krause, G. L., and W. T. Wilson. 1981. Honey bee pollination and visitation patterns on hybrid oilseed sunflowers in central Wyoming (Hymenoptera: Apidae). J. Kansas Entomol. Soc. 54: 75–82.

McGregor, S. E. 1976. Insect pollination of cultivated crop plants. USDA Agric. Handbook, No. 496, U.S. Gov. Printing Office, Washington, DC, 411 p.

Mitchell, J. C., and J. F. Petolino. 1988. Heat stress effects on isolated reproductive organs of maize. J. Plant Physiol. 133: 625–628.

Myers, R. L. 2002. Sunflower. A native oilseed with growing markets (www.jeffersoninstitute.org).

Ortega, E., J. Egea, and F. Dicenta. 2004. Effective pollination period in almond cultivars. HortScience 39: 19–22.

Parker, F. D. 1981. Sunflower pollination: abundance, diversity and seasonality of bees and their effect on seed yields. J. Apic. Res. 20: 49–61.

Peet, M. M., S. Sato, and R. Gardener. 1998. Comparing heat stress effects on male- fertile and male-sterile tomatoes. Plant Cell Environ. 21: 225–231.

Postweiler, K., R. Stosser, and S. F. Anveri. 1985. The effect of different temperatures on the viability of ovules in cherries. Sci. Hort. 92: 125–135.

Prasad, P.V.V., P. Q. Crauford, V. G. Kakani, T. R. Wheeler, and K. Boote. 2001. Influence of high temperature during pre- and post-anthesis stages of floral development on fruit-set and pollen germination in peanut. Aust. J. Plant Physiol. 28: 233–240.

Reddy, K. R., H. F. Hodges, and V. R. Reddy. 1992. Temperature effects on cotton fruit retention. Agron. J. 84: 26–30.

Ribbands, C. R. 1953. The behaviour and social life of honeybees. Bee Research Association, London, UK.

Rosell, P., M. Herrero, and V. G. Sauco. 1999. Pollen germination of cherimoya (*Annona cherimmola* Mill.). In vivo characterization and optimization of in vitro germination. Sci. Hort. 81: 251–265.

Sato, S. M., M. Peet, and J. F. Thomas. 2002. Determining critical pre- and post-anthesis periods and physiological processes in *Lycopersicon esculentum* Mill exposed to moderately elevated temperatures. J. Exp. Bot. 53: 1187–1195.

Skinner, J. A. 1987. Abundance and spatial distribution of bees visiting male-sterile and male-fertile sunflower cultivars in California. Environ. Entomol. 16: 922–927.

Sokal, R. R., and F. J. Rohlf. 1995. Biometry. W. H. Freeman and Company, New York.

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